quantitatively. We applied this technique to the analysis of exoprotein from SDSE.
Methods: The SDSE strains used in this study were 1 reference strain H46A and 23 clinical isolates from patients with invasive disease in Japan. The bacteria were cultured in brain heart infusion broth containing 0.3% yeast extract for 16 hours. After bacterial culture supernatant was prepared with TCA-acetone concentration, aliquots of the samples were loaded onto 7cm Immobiline DryStrip gels (pH 3 to 10). The first-dimensional electrophoresis conditions and second-dimensional SDS-PAGE separation were performed. The gels were stained with coomassie brilliant blue. Those proteins in the spots were also identified by peptide mass mapping analysis.
Results: We identified 25 spots of exoprotein in SDSE H46A strain by 2-DE analysis. Furthermore we found various amount of Streptokinase among 23 clinical strains by 2-DE analysis and we could also divide 3 exoprotein patterns by the production of Streptokinase in 24 SDSE.
Conclusion: Our study suggests that the 2-DE analysis is useful for categorizing the exoprotein patterns, especially Streptokinase, in SDSE strains.

OL-016 The turn-around-times (TATs) of BacT/ALERT 3D™ and VersaTREK™

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We compared the Turn-Around-Times (TATs) of two competing automated microbial detection systems BacT/ALERT 3D™ (BACT) (bioMérieux, Inc., Durham, NC) and the VersaTREK™ (VTRK) (TREK Diagnostic Systems, Cleveland, OH) through the blood culture analysis results of the Armed Forces of the Philippines Medical Center (Quezon City, Philippines) from January-December 2010. Blood cultures were positive at the rate of 182/888 (25.78%) for BTAC and 159/898 (21.54%) for VTRK. The positivity rate for the two systems was not significantly different ($X^2 = 2.2151$, df = 1, p = 0.1367). Organisms that grew in the blood culture bottles were identified as *Staphylococcus aureus* (n = 36), *Staphylococcus epidermidis (n = 119), Enterobacter aerogenes* (n = 36), *Enterobacter agglomerans* (n = 3), *Enterobacter cloaceae* (n = 5), *Escherichia coli* (n = 21), *Proteus spp* (n = 21), *Acinetobacter spp (n = 61), Alcaligenes spp.* (n = 24) *and Pseudomonas aeruginosa* (n = 6).
Our data revealed the TAT for BTAC to detect blood pathogens is 23.69±14.85 hours and 22.24±12.77 hours for VTRK. The difference between the two systems was not significant (z = 0.9734, p-value = 0.3304). The TAT of systems is not affected by the strain type of organism detected (F = 1.6988, df = 9, p = 0.0882). On the otherhand, methicillin resistance in *S. aureus* and *S. epidermidis* did affect the TAT of detection (F = 0.0806, df = 1, p = 0.7782 and F = 1.9329, df = 1, p = 0.161 respectively).
From our findings, we conclude that the BTAC and VTRK systems are comparable in terms of the positivity rate and TAT for the detection of bloodstream bacterial infections. Likewise, the antibiotic resistance of the organisms did affect the TAT of the systems.

Free Paper Presentation 3: Travel Medicine, Vector-Borne Diseases and Parasitic Infections

Friday, July 15, 2011, 15:30–17:00
Meeting Room 311B

PL-003 Seroepidemiology of *Anaplasma phagocytophilum* among farm worker populations in the Tianjin area during 2007–2009

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Objective: Spotted fever, Human granulocytic anaplasmosis (HGA) and monocytic ehrlichiosis (HME) are worldwide tick-borne rickettsial diseases (TBRD) caused by the obligate intracellular bacteria spotted fever group rickettsiae (SFGR), *Anaplasma phagocytophilum* and *Ehrlichia chaffeensis* respectively. In 2006, the seroepidemiologic data from Tianjin area demonstrated average infection prevalences in farm population as 8.8% similar to rates among serosurveys in North America and Europe. In this study, we describe a continuous seroprevalence investigation of *A. phagocytophilum* in Tianjin areas from 2007 to 2009.
Methods: Field epidemiological surveys were performed in 8 districts of Tianjin and 886 farmers were randomly recruited and their serum samples were collected to detect the specific IgG antibodies of *A. phagocytophilum* by micro-indirect immunoflorescence (IFA).
Results: The IgG antibody positive rates of *A. phagocytophilum* increased from 8.8% in 2006 to 59.2% in 2009 while *E. chaffeensis had an* increase from 0.0% in 2006 to 4.4% in 2009. However, spotted fever group rickettsiae decreased from 1.6% in 2007 to 0.0% in 2009.
Conclusion: Infections of both *A. phagocytophilum* and *E. chaffeensis* in farmers from Tianjin areas were popular and the antibody positive rates of *A. phagocytophilum* and *E. chaffeensis* increased annually. Differential diagnosis for ricketssial diseases in clinical practice and watch out for outbreaks of anaplasmosis and ehrlichiosis in rural areas should be emphasized. Further investigation on vectors and hosts of these ricketssioses in Tianjin areas should also be performed.
Supported by the special project of key communicable viral hepatitis-research on infectious diseases surveillance platform of national Sci-Tech key project (2009ZX10004–203); and on pathogen laboratory network surveillance technique (2008ZX10004–008); and the national basic research project-973 plan (2010CB530206).

OL-017 Aquaporin water channel AgAQP1 in the malaria vector mosquito *Anopheles gambiae* during blood feeding and humidity adaptation

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Background: Altered patterns of malaria endemicity reflect changes in the feeding behavior and climate adaptation of Anopheles gambiae mosquito, which require fast transmembrane water movement. We anticipate that AQP water channels play important role in the physiology of this malaria mosquito vector.

Methods: Genome sequences were analyzed and alignments were performed. Oocyte swelling assay was utilized for in vitro characterization. A desiccation assay and immunofluorescence microscopy were performed to investigate its role in mosquitoes.
Result: We have cloned A. gambiae aquaporin 1 (AgAQP1), which is homologous to known AQPs in humans, Drosophila, and sap-sucking insects. Expressed in Xenopous oocytes, AgAQP1 transports water but not glycerol. Similar to mammalian AQPs, water permeation of AgAQP1 is inhibited by HgCl2 and tetraethylammonium, with Tyr185 conferring tetraethylammonium sensitivity. AgAQP1 is more highly expressed in adult female *A. gambiae* mosquitoes than in males, also high in gut, ovaries, and Malpighian tubules where immunofluorescence microscopy reveals that AgAQP1 resides in stellate cells but not principal cells. AgAQP1 expression is up-regulated in fat body and ovary by blood feeding but not by sugar feeding, and it is reduced by exposure to a dehydrating environment (42% relative humidity). RNA interference reduces AgAQP1 mRNA and protein levels. In a desiccating environment (<20% relative humidity), mosquitoes with reduced AgAQP1 protein survive significantly longer than controls.
Conclusion: These studies support a role for AgAQP1 in water homeostasis during blood feeding and humidity adaptation of A. gambiae, a major mosquito vector of human malaria in sub-Saharan Africa.

OL-018 A rapid way to titrating Hantaan virus with flow cytometry method

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Objective: Using flow cytometry method to detect and titrate Hantaan virus.
Methods: Vero-E6 cells were infected with Hantaan virus 76–118 strain. The Hantaan virus nucleoprotein antigens were detected with monoclonal antibody 3G1 and FITC-labled secondary antibody. After incubation for 12 h, 24 h, 36 h, 48 h and 72 h at 37°C in 5% CO_2, the samples were analyzed on a FACScan flow cytometer to measure the percentage of positive cells. The results were also compared with the indirect immunofluorescence assay.
Results: We found that the optimal time point to measure virus titer was at 36 h after infection, and the percentage of the positive cells was (10.06±0.42) %. The infectious unit determined by FCM was 1.9×10^6/mL, and the lower limit of FCM method was 47.5 IU/mL.
Conclusion: Compared to the classical plaque assay and IFA method, FCM is a rapid and simple way to detect and titrate Hantaan virus.

OL-019 Development of a rapid, sensitive and specific LAMP for detecting *Anaplasma phagocytophilum*

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Background: *Anaplasma phagocytophilum* is the casusative agent of human granulocytic anaplasmosis which national wide prevalent in China. In this study, we develop a rapid, simple and sensitive loop-mediated isothermal amplification (LAMP) assay for detecting *A. phagocytophilum*, which is especially for application in rural areas in China.
Method: A primer sets targeting on *msp2* gene was designed after aligning 82 sequences of *A. phagocytophilum* obtained from Genbank. A recombinant plasmid was constructed as references to analyze the sensitive and repetitive of LAMP assay. 24 members of order rickettsiales and 15 common clinical pathogens were used to define the specificity of LAMP. The developed LAMP in the study, previously established nested PCR and real-time PCR were used to detect 15 blood DNA samples from acute-phase of confirmed cases with anaplasmosis serologically and 27 probable cases.
Result: The sensitivity of LAMP was 100 copies per reaction (25μl) and its specificity was 100%. 26 of 42 samples (15 confirmed cases and 27 probable cases) reacted positively by LAMP within 1 hour, while only 1 and 3 detected by nested PCR and real-time PCR, respectively.
Conclusion: The developed LAMP in the study showed a high sensitivity comparable to that of the nested PCR and real-time PCR for the detection of *A. phagocytophilum*. This LAMP assay is a valuable method for its rapid, cost-effective and simple detection for detecting *A. phagocytophilum* in rural areas of China.
Acknowledgements: This study was funded by National Basic Research Program of China (973 Program) 2010CB530200 (2010CB530206); the National Key Science and Technology Projects of China (No. 2009ZX10004–203) and (No.2008ZX10004–008).

OL-020 Serological investigation of *Rickettsia typhi* on farmers and domestic animals in Yunnan Province, China

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Objectives: Epidemic typhus caused by *R. prowazekii* and endemic typhus caused by *R. typhi* have been listed the second degree infectious diseases according to the law of control and prevention for infectious diseases since the new People's Republic of China in 1949. Local outbreaks of endemic typhus have not been suspended although outbreaks of epidemic typhus are rare in China. Total three larger outbreaks of endemic typhus, which involved in ten thousand peoples, happened since new China was established and each involved in Yunnan Province because of high density of wild mouse in this areas. However, systemic epidemic data on endemic typhus are very limited. In order to investigate the seroprevalence of *R. typhi*, we conducted field survey of *R. typhi* on farmer population and domestic animals during March 10–25, 2009.
Methods: A total of 237 adult farmer sera (aged from 36 to 66), 81 children sera (aged from 4 to 6 years old) and 270 animal sera (90 dogs, 90 ox and 90 goats) were collected from Xundian Country, Yulong Country and Simao Country. IgM and IgG antibodies of *R. typhi* were examined by using immunofluorescence assays (IFA). Samples reactive above the 1:80 and 1:40 screening dilution were deemed positive.
Results: The total IgG seroepidemiologic rates of *R. typhi* for adult farmers was 16.46% and that of children was 12.35%. The total IgG positive rate of *R. typhi* for 3 species of animals was 61.48%. Among these animals, the goats shared the highest IgG positive rates of *R. typhi* (75.76%). Similar seroepidemiologic features were found in the results of IgM antibodies assays.
Conclusion: *R. typhi* infection of farm population and domestic animals are common in Yunnan Province, and